

# **TOXICITY OF PHOSPHINE AT VARIOUS CONCENTRATIONS AND TEMPERATURES TO THE CIGARETTE BEETLE**

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# TOXICITY OF PHOSPHINE AT VARIOUS CONCENTRATIONS AND TEMPERATURES TO THE CIGARETTE BEETLE

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## SUMMARY

Phosphine ( $\text{PH}_3$ ) was tested as a fumigant against all life stages of the cigarette beetle, *Lasioderma serricorne* (F.), in 8-cu.-ft. fumatoriums. Eggs, pupae, larvae, and adults were decreasingly resistant to  $\text{PH}_3$ . Exposure of 72 hours at 80° F. was sufficient to kill all insects in all stages with  $\text{PH}_3$  concentrations of 100, 200, 300, 400, or 500 p.p.m. A concentration of 100 p.p.m. was nearly as effective as 200, 300, 400, or 500 p.p.m. at 80° F. Overall, however, it appears that a concentration of 200 p.p.m. is preferred because higher concentrations did not increase the rate of insect mortality. Effective fumigation of tobacco with  $\text{PH}_3$  may be anticipated when the temperature of the commodity is above 68° F. and the fumigation period is not less than 96 hours. At 60° F. or lower, some eggs and pupae will survive. The suggested fumigation time at 60° F. is 144 hours; at 40° F. it is 240 hours.

## INTRODUCTION

The most destructive insect pest of stored tobacco in the United States is the cigarette beetle, *Lasioderma serricorne* (F.). Phosphine ( $\text{PH}_3$ ) has been used since 1969 in the United States as a space fumigant for control of insects infesting tobacco bales, cases, and hogsheads. Lindgren and Vincent (8)<sup>3</sup> were among the first to report

that low concentrations of  $\text{PH}_3$  were effective against the adult and larval stages of the cigarette beetle. Later, Childs et al. (2, 3) found that  $\text{PH}_3$  readily penetrated tobacco hogsheads; as a space fumigant, it was effective for control of the cigarette beetle in tobacco storages. Dieterich et al. (4) concluded that  $\text{PH}_3$  residue remaining in several types of stored products was generally less than 0.01 p.p.m. after fumigation and a brief period of aeration. These and other investigators have found that  $\text{PH}_3$  is an effective fumigant against most insect pests of stored products.

Few studies have been made to ascertain the effectiveness of  $\text{PH}_3$  at various temperatures and concentrations against each stage of an insect. We made the tests reported here to determine the minimum time required to kill each stage of the cigarette beetle by exposure to  $\text{PH}_3$  concentrations of approximately 100, 200, 300, 400, or 500 p.p.m. at 80° F. In addition, we evaluated the efficacy of  $\text{PH}_3$  at 500 p.p.m. in fumigations at 40°, 60°, 68°, and 80° F.

Brown and Reynolds (1) proposed that 100% kill of insects cannot be estimated from observations of dosage-mortality responses. Drawing from observations of methyl bromide fumigations, a dosage extrapolated to give complete kill of a finite population, if increased would give complete kill of a bag consignment of produce treatments. In contrast, Höglund (5) found that if the dosage is calculated to kill 99% of the resistant insect stage, the calculated increase factor is more than enough to kill the insects in a properly fumigated bag. In our experiment, the dosage was changed to give a range of 100% kill. The calculated, or estimated, dose required to kill 99% of the insects in a

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<sup>3</sup> Italicized numbers in parentheses refer to items in "Literature Cited," p. 7.

of the cigarette beetle may be used as a reference by the fumigator to determine how long to hold the fumigant to kill all stages of the insect infesting the commodity.

## EXPERIMENTAL PROCEDURE

The fumigation of cigarette beetles with PH<sub>3</sub> was conducted in two fumatoriums, each with inside dimensions of 2 ft. by 2 ft. by 2 ft. The fumatoriums were made from sheets of  $\frac{1}{4}$ -inch, transparent, Lucite<sup>4</sup> plastic. One access port, 1 inch in diameter, was in the center of the top wall; another was in a side wall. During fumigation these ports were closed with rubber stoppers. The fumatoriums were housed in a 200-cu.-ft. chamber with controlled temperature and 60%  $\pm$  2% relative humidity.

Phosphine was generated from decomposition of an aluminum phosphide powder composed of 55% aluminum phosphide, 41% ammonium carbamate, and 4% paraffin. In the air space of an 8-cu.-ft. fumatorium, 105 mg. of powder liberated approximately 100 p.p.m. of PH<sub>3</sub>. The powder was weighed and placed in a fumatorium 24 to 72 hours before exposure of insects. Just before introduction of the insects, air samples were taken for analysis by gas chromatography (5) to determine the PH<sub>3</sub> concentration in the fumatoriums. Thereafter, air samples were taken every 24 hours with detector tubes (6) to check for changes in fumigant concentrations. (During some fumigations a limited number of air samples were analyzed by gas chromatography to confirm results taken with the detector tubes.) After each fumigation, air was passed through the chambers for 3 days before another test was started.

The approximate ages at which each stage of the cigarette beetle is most resistant to PH<sub>3</sub> were determined as follows: eggs—0 to 1 day old; larvae—4 to 5 days in the fourth instar; pupae—1 to 2 days old; and adults—2 to 3 days after emergence from the pupal case. Care was taken to always test the most resistant age of each stage.

<sup>4</sup> Trade names are used in this publication solely for the purpose of providing specific information. Mention of a trade name does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture or an endorsement by the Department over other products not mentioned.

The insects were reared on wheat flour in an insectary maintained at 80°  $\pm$  1° F. and 70%  $\pm$  2% relative humidity. The beetle oviposited on  $\frac{3}{4}$ -inch sections of flue-cured tobacco stems that had previously been split. Adults, larvae, and pupae exposed to PH<sub>3</sub> at 40° F. were preconditioned for 72 hours at 60° F. before exposure in the fumatoriums.

The insects were exposed in cages  $\frac{3}{4}$ -inch in diameter by 2 inches long. The cages, capped with a neoprene stopper, were constructed from 40-mesh, stainless-steel, wire gauze. Larvae and adults were unprotected, but eggs and pupae were protected in the cages against physical injury by exposure in either tobacco stems or pupal cases. Approximately 25 insects of one stage were exposed in each cage. The cages were suspended on a cord in the air space of the fumatoriums. Three cages of an insect stage were removed at a time from a fumatorium through the top access hole at predetermined intervals during fumigation. The length of exposure for each insect stage depended on its resistance to the fumigant.

Lengths of exposures were selected to establish four points of mortality below 90% and four points from 90% to 100%, except for fumigations at 68° F. An additional exposure expected to be long enough to consistently kill 100% of the insects was scheduled. All exposures that killed less than 100% of the insects were replicated 3 times (3 cages of insects  $\times$  25 insects per cage  $\times$  3 replications, for a total of 225 insects of a stage for each exposure period). The exposure that killed 100% of the insects was replicated 6 times. At 68° F. only those exposures that killed 90% to 100% of the insects and those that consistently killed all insects of a stage were evaluated.

Three cages of insects of each stage served as controls for each replicated fumigation. These insects were held in the controlled temperature-humidity room that housed the fumatoriums. The insects remained in this room for the same time required to kill all insects of an identical stage exposed in the fumatoriums.

The mortality of cigarette beetle adults and larvae was recorded after a postexposure period of 3 days; of pupae, after 11 days; and of eggs, after 9 and 12 days. Larvae from eggs that were first to hatch were removed from the tobacco

stems after 9 days to reduce the possibility of cannibalism. The insects during the period of postexposure were held in a room maintained at  $80^\circ \pm 2^\circ$  F. and  $70\% \pm 2\%$  relative humidity.

Moribund insects were recorded as alive. The mortality of pupae was decided by the number of dead pupae in pupal cases after a postexposure period of 11 days. Eggs that had not hatched 12 days after fumigation were considered dead.

The results of this experiment are reported as estimated and observed insect mortality. The statistical methods followed in the calculation of estimated mortalities are discussed in the appendix.

## RESULTS AND DISCUSSION

The data reported in this experiment resulted from the fumigations reported in table 1. In all fumigations the actual concentration of PH<sub>3</sub> in the fumatoriums was close to the planned concen-

tration. Fumigations below 80° F. deviated more from the planned concentrations than those at 80° F. The average loss of PH<sub>3</sub> during fumigation was approximately 20 p.p.m. and never exceeded 40 p.p.m. The greatest fumigant loss occurred at 40° F. and with exposures of several days.

The average mortality of control insects in tests at 80° F. and planned PH<sub>3</sub> concentrations of 100, 200, 300, 400, or 500 p.p.m. was less than 1% for adults and larvae, approximately 2% for eggs, and 3% for pupae.

### Susceptibility of Insects in Different Growth Stages

LD<sub>50</sub> values, obtained by logarithmic probit analysis, were used to compare the susceptibility of the insect stages to PH<sub>3</sub>. The calculated LD<sub>50</sub> values of each insect stage were generally in close agreement with the observed exposure

TABLE 1.—Planned and actual phosphine concentrations in fumatoriums at beginning of each replication of a test condition

| Temperature | Replication | Planned |        | Temperature | Replication | Planned |        |
|-------------|-------------|---------|--------|-------------|-------------|---------|--------|
|             |             | P.p.m.  | P.p.m. |             |             | P.p.m.  | P.p.m. |
| 40° F.      | 1           | 500     | 500    | 80° F.      | 1           | 100     | 100    |
|             | 2           | 500     | 500    |             | 2           | 100     | 100    |
|             | 3           | 500     | 480    |             | 3           | 100     | 100    |
|             | 4           | 500     | 500    |             | 4           | 100     | 100    |
|             | 5           | 500     | 500    |             | 5           | 100     | 100    |
|             | 6           | 500     | 508    |             | 6           | 100     | 100    |
| 60° F.      | 1           | 500     | 475    | 80° F.      | 1           | 200     | 195    |
|             | 2           | 500     | 463    |             | 2           | 200     | 194    |
|             | 3           | 500     | 475    |             | 3           | 200     | 185    |
|             | 4           | 500     | 527    |             | 4           | 200     | 185    |
|             | 5           | 500     | 505    |             | 5           | 200     | 185    |
|             | 6           | 500     | 516    |             | 6           | 200     | 196    |
| 68° F.      | 1           | 500     | 555    | 80° F.      | 1           | 300     | 307    |
|             | 2           | 500     | 537    |             | 2           | 300     | 319    |
|             | 3           | 500     | 575    |             | 3           | 300     | 330    |
|             | 4           | 500     | 460    |             | 4           | 300     | 288    |
|             | 5           | 500     | 477    |             | 5           | 300     | 288    |
|             | 6           | 500     | 511    |             | 6           | 300     | 288    |
| 80° F.      | 1           | 500     | 500    | 80° F.      | 1           | 400     | 390    |
|             | 2           | 500     | 518    |             | 2           | 400     | 408    |
|             | 3           | 500     | 525    |             | 3           | 400     | 390    |
|             | 4           | 500     | 525    |             | 4           | 400     | 400    |
|             | 5           | 500     | 525    |             | 5           | 400     | 400    |
|             | 6           | 500     | 502    |             | 6           | 400     | 400    |

TABLE 2.—Calculated<sup>1</sup> and observed length of exposure required to kill 50% of insects exposed to 500 p.p.m. of phosphine

| Method of tabulation | Hours insects exposed to phosphine at temperatures (F.) of— |     |                   |     |                   |        |  |
|----------------------|---|-----|-------------------|-----|-------------------|--------|--|
|                      | 40°   | 60° | 80°               | 40° | 60°               | 80°    |  |
| Adults               |   |     |                   |     |                   | Larvae |  |
| Calculated           | 5.5   | 3.4 | 0.7               | 6.1 | 9.5               | 7.1    |  |
| Observed             | 6.0   | 4.0 | .8                | 6.0 | <sup>2</sup> 16.0 | 8.0    |  |
| Pupae                |   |     |                   |     |                   | Eggs   |  |
| Calculated           | 26  | 27  | 7.1               | 54  | 58                | 19     |  |
| Observed             | 32  | 32  | <sup>3</sup> 16.0 | 56  | 96                | 18     |  |

<sup>1</sup> LD<sub>50</sub> as determined by logarithmic probit analysis.

<sup>2</sup> At 6 hours, observed mortality was 49.5%. No observations were made between 6 and 16 hours.

<sup>3</sup> At 8 hours, observed mortality was 46.7%. No observations were made between 8 and 16 hours.

hours required for 50% or greater kill (table 2). Few mortality readings were taken for larvae and pupae exposed to PH<sub>3</sub> for less than 24 hours. As a result, the apparent lack of agreement between calculated and observed hours for 50% mortality of larvae at 60° F. and pupae at 80° F. is exaggerated in table 2.

#### Tests at Five Phosphine Levels

At PH<sub>3</sub> concentrations of 100, 200, 300, 400, or 500 p.p.m. and 80° F., the egg is the most phosphine-resistant stage of the cigarette beetle (fig.

1). The adult is the least resistant. Pupal and larval stages have intermediate resistance.

A concentration of 100 p.p.m. of PH<sub>3</sub> was nearly as effective as 500 p.p.m. at 80° F. in killing the beetle. Overall, however, it appears that a concentration of 200 p.p.m. is preferred because greater concentrations require nearly the same duration of exposure to kill an equal number of insects. The egg was more susceptible to the gas at 200 p.p.m. (14.1 hours of exposure for 50% kill) than at 100 p.p.m. (24.8 hours of exposure).

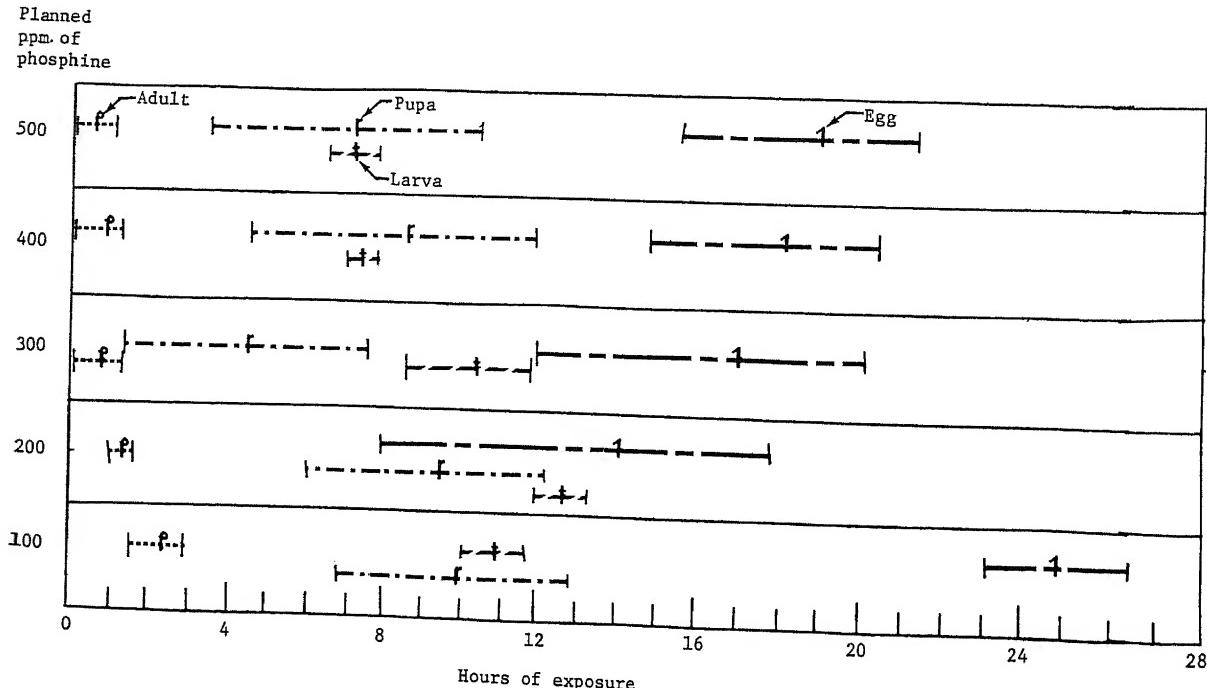


Figure 1.—Estimated hours of phosphine exposure required to kill 50% of cigarette beetles at 80° F. (upper and lower limits; 95% confidence for LD<sub>50</sub> values).

### Tests at Three Exposure Temperatures

The effect of temperature on the efficiency of PH<sub>3</sub> for control of the cigarette beetle is shown in figure 2. All stages of the cigarette beetle were killed in shorter exposures at 80° F. than at either 60° or 40° F. Egg mortality of 50% occurred after 19 hours at 80° F. but 58 and 54 hours were required for the same degree of kill at 60° and 40° F., respectively. The susceptibility of larvae and pupae was similar at 80° F., but at 60° and 40° F. the pupal stage was more difficult to kill and its resistance approached that of the egg. Adults required progressively longer times for 50% kill as temperature was lowered from 80° to 60° to 40° F., but in all cases 50% kill occurred in 5.5 hours or less.

### Hours of Fumigation Required For Control of the Cigarette Beetle

LD<sub>50</sub> values obtained by logarithmic analysis and results of fumigations of sufficient duration to kill all insects were used as indicators of the length of time required for complete control of the cigarette beetle.

### Fumigation at 80° F.

Exposure of 72 hours at 80° F. was sufficient to kill all insects in all growth stages with PH<sub>3</sub> concentrations of 100 to 500 p.p.m. (table 3). The estimated exposures indicate that some survival of pupae and eggs might be expected if the testing were repeated over and over. However, these data signify that such survival would be infrequent. There is probably no benefit to be derived from going above a 200-p.p.m. fumigant concentration.

### Fumigation at Low Temperatures

The effectiveness of PH<sub>3</sub> against the cigarette beetle declined with temperature (table 4). PH<sub>3</sub> at 500 p.p.m. killed the insects in all stages after exposure times of 40 hours at 80° F., 96 hours at 68° F., and 144 hours at 60° F. At 40° F. some pupae survived even 480 hours of fumigation.

The susceptibility of the adult and larval stages to PH<sub>3</sub> decreased when fumigations were below 80° F. The susceptibility of the larval stage, however, was similar at 40°, 60°, and 68° F. The eggs and pupae were much more resist-

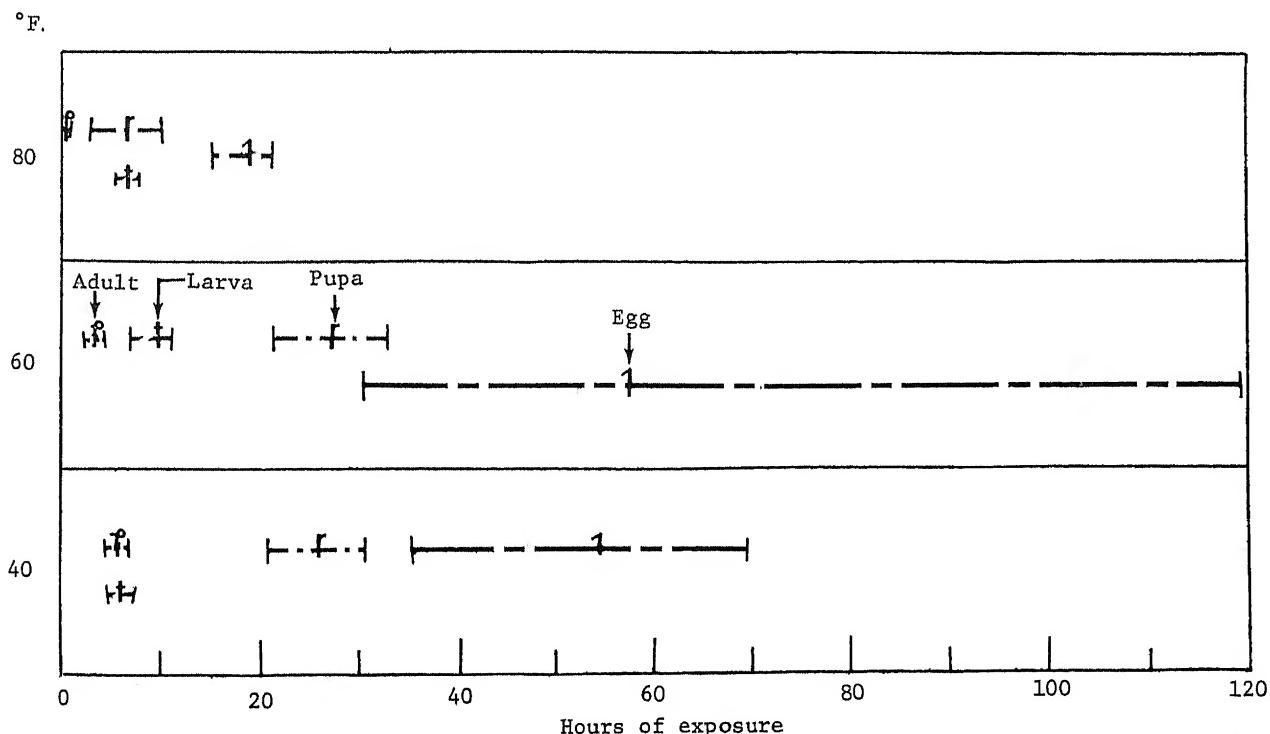


Figure 2.—Estimated hours of exposure to 500 p.p.m. of phosphine required to kill 50% of cigarette beetles at 40°, 60°, and 80° F. (upper and lower limits; 95% confidence for LD<sub>50</sub> values).

ant to PH<sub>3</sub> below 80° F. At 60° F. an exposure of 144 hours at 500 p.p.m. was needed to kill all eggs and pupae. At 40° F. all eggs were killed in 240 hours, but some pupae survived even after 480 hours. It was estimated that an exposure of 1,714 hours at 500 p.p.m. PH<sub>3</sub> would be needed to kill 99% of the pupae at 40° F. The mortality of eggs fumigated 240 hours at 40° F. appeared to be due nearly as much to the low temperature as to the PH<sub>3</sub>. Egg mortality of control insects at 40° F. was 98%.

These data indicate that the fumigation period

should be extended when commodities with a temperature lower than 80° F. are fumigated with PH<sub>3</sub> for control of the cigarette beetle. Effective fumigation of tobacco with PH<sub>3</sub> may be anticipated when the commodity is above 68° F. and the fumigation period is not less than 96 hours. At 60° F. or lower, some eggs and pupae may be expected to survive. If fumigation is conducted at these low temperatures, the suggested fumigation times are 144 hours at 60° F. and 240 hours at 40° F.

TABLE 3.—Estimated and observed exposures needed to kill each stage of the cigarette beetle exposed to phosphine concentrations of 100, 200, 300, 400, or 500 p.p.m. at 80° F.

| Insect stage | Phosphine | Estimated exposures |  | Observed exposures |         | 100% mortality |
|--------------|-----------|---------------------|--|--------------------|---------|----------------|
|              |           | LD <sub>99</sub>    | Upper limit <sup>1</sup><br>LD <sub>99</sub> | Cages <sup>2</sup> | Insects |                |
| Adult        | P.p.m.    | Hours               | Hours  | Number             | Number  | Hours          |
|              | 100       | 6.2                 | 17.0   | 27                 | 680     | 16             |
|              | 200       | 4.1                 | 5.2  | 18                 | 441     | 16             |
|              | 300       | 6.8                 | 46.1   | 18                 | 461     | 16             |
|              | 400       | 3.4                 | 41.5   | 24                 | 604     | 16             |
| Larval       | 500       | 2.8                 | 10.2   | 24                 | 615     | 16             |
|              | 100       | 40.3                | 47.8   | 18                 | 377     | 48             |
|              | 200       | 39.8                | 43.3   | 21                 | 528     | 48             |
|              | 300       | 46.3                | 60.2   | 18                 | 450     | 44             |
|              | 400       | 33.8                | 37.4   | 27                 | 680     | 40             |
| Pupal        | 500       | 37.6                | 46.0   | 24                 | 600     | 40             |
|              | 100       | 96.0                | 191.0  | 27                 | 597     | 56             |
|              | 200       | 70.1                | 139.0  | 21                 | 562     | 56             |
|              | 300       | 101.0               | 794.0  | 21                 | 596     | 44             |
|              | 400       | 101.0               | 397.0  | 24                 | 583     | 44             |
| Egg          | 500       | 69.3                | 268.0  | 24                 | 598     | 40             |
|              | 100       | 44.1                | 51.8   | 60                 | 1,712   | 72             |
|              | 200       | 51.1                | 103.0  | 24                 | 429     | 44             |
|              | 300       | 53.3                | 88.7   | 24                 | 403     | 44             |
|              | 400       | 50.9                | 80.6   | 24                 | 392     | 44             |
|              | 500       | 51.3                | 71.1   | 21                 | 360     | 40             |

<sup>1</sup> 95% confidence limit for LD<sub>99</sub> value.

<sup>2</sup> Each cage contained approximately 25 insects. Exposures were replicated 6 times with each replication consisting of a new fumigation and of tests of 3 or more cages.

TABLE 4.—Estimated and observed exposures needed to kill each stage of the cigarette beetle exposed to 500 p.p.m. of phosphine at 40°, 60°, 68°, or 80° F.

| Temperature<br>and<br>insect stage | Estimated exposures       |   | Observed exposures |         | 100 %<br>mortality<br>Hours | Mortality in<br>absence of PH <sub>3</sub><br>% |
|------------------------------------|---------------------------|---|--------------------|---------|-----------------------------|---|
|                                    | LD <sub>99</sub><br>Hours | Upper limit <sup>1</sup><br>LD <sub>99</sub><br>Hours | Cages <sup>2</sup> | Insects |                             |   |
| <b>40° F.:</b>                     |                           |   |                    |         |                             |   |
| Adult                              | 18.0                      | 35.1  | 59                 | 1,489   | 40                          | 1.7   |
| Larval                             | 51.0                      | 67.3  | 30                 | 721     | 56                          | 8.3   |
| Pupal                              | 1,714.0                   | 3,122.0   | (3)                | (3)     | (3)                         | (3)   |
| Egg                                | 305.0                     | 627.0   | 30                 | 630     | 240                         | 93.0  |
| <b>60° F.:</b>                     |                           |   |                    |         |                             |   |
| Adult                              | 9.7                       | 14.8  | 24                 | 626     | 24                          | 1.2   |
| Larval                             | 65.1                      | 89.4  | 33                 | 856     | 72                          | 6.1   |
| Pupal                              | 276.0                     | 538.0   | 18                 | 436     | 144                         | 1.8   |
| Egg                                | 672.0                     | —   | 18                 | 414     | 144                         | 10.7  |
| <b>68° F.:</b>                     |                           |   |                    |         |                             |   |
| Adult                              | —                         | —   | 45                 | 1,177   | 24                          | .7  |
| Larval                             | —                         | —   | 45                 | 1,139   | 76                          | 3.5   |
| Pupal                              | —                         | —   | 57                 | 1,331   | 96                          | .3  |
| Egg                                | —                         | —   | 21                 | 456     | 96                          | 5.7   |
| <b>80° F.:</b>                     |                           |   |                    |         |                             |   |
| Adult                              | 2.8                       | 10.2  | 24                 | 615     | 16                          | .1  |
| Larval                             | 37.6                      | 46.0  | 24                 | 600     | 40                          | .3  |
| Pupal                              | 69.3                      | 268.0   | 24                 | 598     | 40                          | 2.4   |
| Egg                                | 51.3                      | 71.1  | 21                 | 360     | 40                          | .8  |

<sup>1</sup> 95% confidence limit for LD<sub>99</sub> value.

<sup>2</sup> Each cage contained approximately 25 insects. Exposures were replicated 6 times with each replication consisting of new fumigation and of tests of 3 or more cages.

<sup>3</sup> Not determined; greater than 480 hours.

## LITERATURE CITED

- (1) Brown, W. B., and Reynolds, E. M. 1964. FUMIGATION AIMED AT COMPLETE MORTALITY. 12th Internat. Cong. Ent. Proc. London, 1964: 656.
- (2) Childs, D. P., Overby, J. E., and Niffenegger, D. 1969. PHOSPHINE FUMIGATION OF FLUE-CURED TOBACCO WAREHOUSES FOR CONTROL OF THE CIGARETTE BEETLE. *Tobacco Sci.* 13: 64-69.
- (3) ——— and Watkins, B. J. 1969. PHOSPHINE FUMIGATION OF FLUE-CURED TOBACCO FOR CONTROL OF THE CIGARETTE BEETLE: LABORATORY TESTS. *Tobacco Sci.* 13: 160-163.
- (4) Dieterich, W. H., Mayr, G., Hild, K., and others. 1967. HYDROGEN PHOSPHIDE AS A FUMIGANT FOR FOODS, FEEDS, AND PROCESSED FOOD PRODUCTS. *Residue Rev.* 19: 135-149.
- (5) Dumas, Theodore. 1964. DETERMINATION OF PHOSPHIDE IN AIR BY GAS CHROMATOGRAPHY. *Agr. and Food Chem.* 12(8): 257-258.
- (6) ——— and Monro, H. A. U. 1966. DETECTOR TUBES FOR FIELD DETERMINATION OF FUMIGANT CONCENTRATIONS. *Pest Control* 7(34): 20-32, 52.
- (7) Howe, R. W., and Hole, B. D. 1967. PREDICTING THE DOSAGE OF FUMIGANT NEEDED TO ERADICATE INSECT PESTS FROM STORED PRODUCTS. *Jour. Appl. Ecol.* 4: 337-351.
- (8) Lindgren, D. L., and Vincent, L. E. 1966. RELATIVE TOXICITY OF HYDROGEN PHOSPHIDE TO VARIOUS STORED-PRODUCT INSECTS. *Jour. Stored Prod. Res.* 2: 141-146.

## APPENDIX.—STATISTICAL CALCULATIONS

This experiment produced typical dosage-mortality data—that is, the data were percentages of dead insects observed after various time intervals in replicated fumigation tests. Average mortality percentages were calculated for each insect stage for each combination of dosage rate and hours of fumigation. Graphs indicated that an asymptotic regression equation might be fitted to the data. This was done. In most cases, these curves agreed quite well with plots of the observed mortality percentages. Predicted numbers of hours required to reach 90%, 95%, 99%, and 99.9% mortality appeared quite reasonable. However, there were occasions when some asymptotes were greater than 100% (one as high as 185%). We removed this problem by recalculating the equations so the asymptote was not allowed to go above 100%. This procedure, however, gave less than satisfactory answers with some data sets that had responded well in the earlier calculations.

Next we moved to probit analyses. Both arithmetic and logarithmic probit analyses were applied to all data sets. Some data sets responded better to the arithmetic analyses; other sets made more sense when the logarithmic probit procedure was used.

All data sets gave what appeared to be reasonable answers when they were analyzed by one or more of these curve-fitting procedures. However, we felt that the same analysis should be applied to all data. We settled on the logarithmic probit procedure because it gives reasonable answers most of the time and because it is well

known and widely used.  $LD_{50}$  values looked especially good when they were compared with observed data. Hence,  $LD_{50}$  values, rather than  $LD_{90}$  values, were used for comparing insects in different growth stages for susceptibility to phosphine. While this is of interest from a biological standpoint, practical fumigation requires complete kill rather than 50% kill.

A second type of data was obtained in the experiment: the hours required to kill all insects. These data are recorded in the columns labeled "Observed Exposures" in tables 3 and 4. Such data lend themselves to a type II error argument: What is the probability of some live insects being present in the cigarette beetle population when  $x$  fumigated insects were observed in an experiment and all were dead?

We might solve  $(1-p)^n = Q$  for  $p$  at specified levels of  $Q$  and  $n$ . For example, table 3 shows that 680 adult cigarette beetles were fumigated with 100 p.p.m.  $PH_3$  at 80° F. for 16 hours. All were dead at the end of the fumigation period. Using this information, we can calculate that the true proportion of cigarette beetle adults able to survive this treatment might be as high as 0.676%, and yet one in each 100 experiments so conducted would show zero survival.

The authors welcome comments by others who have wrestled with similar analytical problems. Statistical methodology in this area is at least as uncertain as are the techniques of fumigation application, gas concentration monitoring, insect rearing, etc.

